

Remarks

Entry of the foregoing amendments and the following remarks are respectfully requested.

The specification has been amended to delete blanks which were objected to by Examiner. Entry of this amendment to the specification is respectfully requested.

The specification has also been amended to correct SEQ ID NOs. As is evident from the originally filed application and Sequence Listing, the SEQ ID NO. for LSA-NRC(H) polynucleotide is SEQ ID NO:25 and not SEQ ID NO:22 found in Table 2 on page 12. Similarly for the amino acid sequence of LSA-NRC(H) which is SEQ ID NO:26 as originally filed in the Sequence Listing, and not SEQ ID NO:23 found in Table 2 page 12 as originally filed. Entry of these corrections is respectfully requested.

Claims 1-4, 6-9, 39-42, 44, 46, 49-52, 54-57, 59-62, 64, and 91-93 are being examined.

Claims 5, 10, 11-38, 43, 47, 48, 53, 58, 63, 65-90 have been withdrawn by Examiner as being drawn to a nonelected invention.

Claims 1-3, 6-8, 40-41, 44, 50-51, 54-56, 59-61, 64, 91-92 have been canceled without prejudice in order to expedite prosecution. Applicants retain the right to prosecute the subject matter of the canceled claims in a future divisional or continuation application.

Claims 4, 9, 39, 42, 46, 49, 52, 57, 62, 93 are pending.

Claim 4 has been amended to correct its dependence on a canceled claim. Claim 39 has been amended to correct its multiple dependency on canceled claims and now specifies the polypeptide of claim 4. Claim 49 has been amended to recite the polypeptide of claim 4.

Applicants acknowledge with appreciation the entry and examination of claim 93 drawn to the polypeptide of claim 4 encoded by the polynucleotide specified in SEQ ID NO:25.

Examiner has requested a new Oath/Declaration because the corrections to the previously executed Oath/Declaration were not initialed and dated. An executed, corrected Oath/Declaration will be submitted as soon as it is in hand.

The disclosure stands objected to because of informalities on page 29. A corrected page 29 is submitted herewith. Entry of the corrected page 29 and withdrawal of the objection are respectfully requested.

Claims 91-93 stand rejected under 35 U.S.C. 112, first paragraph as allegedly not enabled. Examiner listed claim 93 drawn to a polynucleotide sequence as part of this rejection. Applicants assume this was an oversight since the Examiner discusses a vaccine in the rejection. Claims 91 and 92 have been canceled. Withdrawal of the rejection is respectfully requested.

Claims 1-4, 6-9, 39-42, 44, 46, 49-52, 54-57, 59-62, 64 and 91-92 stand rejected under 35 U.S.C. 112, first paragraph as allegedly failing to comply with the written description requirements. Claims 3, 8, 39, 44, 54, 59, 64, 91, and 92 stand rejected under 35 U.S.C. 112, second paragraph, as allegedly indefinite. The claims as amended are believed to be definite. Withdrawal of these rejections is respectfully requested.

Claims 1-3, 6-8, 39-41, 44, 50, 51, 54, 55, 56, 59-61, 64 stand rejected under 35 U.S.C. 102(b) as allegedly anticipated by Londono et al. (J. Immunol. 145: 1557-1563, 1990). Claims 1-4, 6-9, 39-42, 44, 46, 49-52, 54-57, 59-62, 64, and 91-93 stand rejected under 35 U.S.C. 102(b) as allegedly anticipated by Guerin-Marchand et al. (US Patent 6,319,502). These rejections are traversed in view of the claim amendments and the following.

The claims as amended are drawn to a polypeptide LSA-NRC(H) specified in SEQ ID NO:26 produced from a polynucleotide which has been harmonized for expression in the host cell. None of the references describe, suggest or make obvious the

In re Application of Lanar et al.
Serial no. 10/706,435

LSA-NRC(H) polypeptide or polynucleotide. Withdrawal of the rejections is respectfully requested.

All objections and rejections have been addressed. This application is believed to be in condition for Allowance and Notice to that effect is respectfully requested. Should the Examiner find the pending claims allowable, Applicants request rejoinder of claims 13 and 16 drawn to a vector containing the polynucleotide sequence of LSA-NRC(H). Should the Examiner have a need to discuss the claims or amendments to the claims, Examiner is encouraged to contact Applicants by telephone.

Respectfully submitted,

By *S. Pratt*, Reg No 39,441
for Elizabeth Arwine
Reg. No. 45,867

U.S. A. MPMC
504 Scott Street
Fort Detrick, MD 21702-5012
ATTN: MCMR -JA (Elizabeth Arwine-Patent Atty)

I hereby certify that this correspondence is being deposited with the United States Postal Service as Express mail with mailing label no. EV905210601US in an envelope addressed to: Mail Stop: Petitions
Commissioner of Patents, P.O. Box 1450, Alexandria, VA 22313-1450, on May 1, 2008.

By *S. Pratt*
Sana A. Pratt



that effective amounts will be found within a relatively large, non-critical range. An appropriate effective amount can be readily determined using only routine experimentation. Preferred ranges of

5 LSA-NRC for prophylaxis of malaria disease are about 0.01 to 1000 ug/dose, more preferably about 0.1 to 100 ug/dose most preferably about 10-50 ug/dose. Several doses may be needed per individual in order to achieve a sufficient immune response and

10 subsequent protection against malaria.

More particularly, the present invention contemplates essentially purified LSA-NRC and a method for isolating or purifying recombinant LSA-NRC protein.

15 The term 'LSA-NRC' refers to a polypeptide or an analogue thereof (e.g. mimotopes) comprising an amino acid sequence (and/or amino acid analogues) defining at least one LSA-1 epitope. Typically, the sequences defining the epitope correspond to the

20 amino acid sequence of LSA-1 region of *P. falciparum* (either identically, by harmonization, or via substitution of analogues of the native amino acid residue that do not destroy the epitope). The LSA-NRC(H) protein or polypeptide corresponds to a

25 nucleotide sequence identified in SEQ ID NO: ~~22~~²⁵ and an amino acid sequence identified in SEQ ID NO: ~~23~~²⁶ which spans from amino acid 28-154 of the N-terminal region, two 17 aa repeats of the 86 possible from the full length molecule, and the C-terminal region

30 amino acids #1630-1909 of LSA-1 3D7 allele. Upon expression in *E. coli* LSA-NRC(H) is expected to have an approximate molecular weight of 53 kDa as determined by SDS-PAGE.



that effective amounts will be found within a relatively large, non-critical range. An appropriate effective amount can be readily determined using only routine experimentation. Preferred ranges of LSA-NRC for prophylaxis of malaria disease are about 0.01 to 1000 ug/dose, more preferably about 0.1 to 100 ug/dose most preferably about 10-50 ug/dose. Several doses may be needed per individual in order to achieve a sufficient immune response and subsequent protection against malaria.

More particularly, the present invention contemplates essentially purified LSA-NRC and a method for isolating or purifying recombinant LSA-NRC protein.

The term 'LSA-NRC' refers to a polypeptide or an analogue thereof (e.g. mimotopes) comprising an amino acid sequence (and/or amino acid analogues) defining at least one LSA-1 epitope. Typically, the sequences defining the epitope correspond to the amino acid sequence of LSA-1 region of *P. falciparum* (either identically, by harmonization, or via substitution of analogues of the native amino acid residue that do not destroy the epitope). The LSA-NRC(H) protein or polypeptide corresponds to a nucleotide sequence identified in SEQ ID NO:25 and an amino acid sequence identified in SEQ ID NO:26 which spans from amino acid 28-154 of the N-terminal region, two 17 aa repeats of the 86 possible from the full length molecule, and the C-terminal region amino acids #1630-1909 of LSA-1 3D7 allele. Upon expression in *E. coli* LSA-NRC(H) is expected to have an approximate molecular weight of 53 kDa as determined by SDS-PAGE.



polypeptides of the present invention are expressed within the cell and are released upon lysing the cells.

It is also understood that the isolates used in the examples section of the present invention were not intended to limit the scope of the invention and that an equivalent sequence from a *P. falciparum* isolate from another allele, e.g. FVO, T9/96 or CAMP, can be used to produce a recombinant LSA-1 protein using the methods described in the present application. Other new strains or clones of *P. falciparum* may be a suitable source of LSA-1 sequence for the practice of the present invention.

The LSA-NRC nucleotide sequence of the present invention can be part of a recombinant vector. Therefore, the present invention relates more particularly to the *lsa-nrc*^{hmut} nucleic acid sequence (SEQ ID NO:3) in recombinant vector, pET KLSA-NRC^{hmut} deposited with ATCC under the Budapest Treaty on _____, and having accession number _____.

The LSA-1 genomic sequence was cloned into the base vector pETK(-) a modified pET32 plasmid vector from Novagen (Madison, Wisconsin). This plasmid comprises, in sequence, a T7 promoter, optionally a lac operator, a ribosome binding site, restriction sites to allow insertion of the structural gene and a T7 terminator sequence. To aid in purification of the expressed protein, a single histidine tag is cloned at the C-terminus. The ampicillin antibiotic resistance gene has been replaced with a kanamycin resistance gene in pETK(-) and the orientation of the kanamycin ORF is opposite to that of the ORF for the gene that is inserted for expression. Examples of other plasmids which contain



polypeptides of the present invention are expressed within the cell and are released upon lysing the cells.

It is also understood that the isolates used in the examples section of the present invention were not intended to limit the scope of the invention and that an equivalent sequence from a *P. falciparum* isolate from another allele, e.g. FVO, T9/96 or CAMP, can be used to produce a recombinant LSA-1 protein using the methods described in the present application. Other new strains or clones of *P. falciparum* may be a suitable source of LSA-1 sequence for the practice of the present invention.

The LSA-NRC nucleotide sequence of the present invention can be part of a recombinant vector. Therefore, the present invention relates more particularly to the *lsa-nrc*^{hmut} nucleic acid sequence (SEQ ID NO:3) in recombinant vector, pET KLSA-NRC^{hmut}. The LSA-1 genomic sequence was cloned into the base vector pETK(-) a modified pET32 plasmid vector from Novagen (Madison, Wisconsin). This plasmid comprises, in sequence, a T7 promoter, optionally a lac operator, a ribosome binding site, restriction sites to allow insertion of the structural gene and a T7 terminator sequence. To aid in purification of the expressed protein, a single histidine tag is cloned at the C-terminus. The ampicillin antibiotic resistance gene has been replaced with a kanamycin resistance gene in pETK(-) and the orientation of the kanamycin ORF is opposite to that of the ORF for the gene that is inserted for expression. Examples of other plasmids which contain